Poly(A) Polymerase, Yeast

CATALOG NO.: M1215-100

AMOUNT: 100 U (100 µl)

PRODUCT SOURCE: Recombinant *E. coli*

FORM: Liquid. Enzyme supplied with 5X Reaction Buffer

COMPONENTS:

Components Name	Volume	Part No.
Poly(A) Polymerase, Yeast (1 U/µl)	100 µl	M1215-100-1
5X Poly(A) Polymerase, Yeast Reaction Buffer	1 ml	M1215-100-2
25 mM $MnCl_2$	500 µl	M1215-100-3
ATP (10 mM)	150 µl	M1215-100-4

DESCRIPTION: Poly(A) Polymerase catalyses the template independent addition of adenosine residues onto the 3' ends of polyribonucleotides. The use of ATP as a substrate leads to poly(A) tailing whereas substitution of cordycepin-5'-triphosphate (3'- dATP) for ATP results in addition of a single dA residue to the 3'-termini of the RNA. Neither ADP nor dATP can be used as substrates for this enzyme. Poly(A) Polymerase from yeast has been shown to be more effective at oligonucleotidelabeling and poly(A) tailing of long RNA templates than Poly(A) Polymerase from E. coli.

APPLICATIONS:
1. Labelling of RNA with ATP or cordycepin
2. Poly(A) tailing of RNA for cloning or affinity purification
3. Increasing translation of RNA transferred into eukaryotic cells

STORAGE CONDITIONS: Store all components at -20°C. Avoid repeated freeze-thaw cycles of all components to retain maximum performance. All components are stable for one year from the date of shipping when stored and handled properly.

ENZYME STORAGE BUFFER: 20 mM Tris-HCl (pH 8.0), 100 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 0.1% Triton® X-100 and 50% (v/v) Glycerol.

ENZYME UNIT DEFINITION: One unit is defined as the amount of Poly(A) Polymerase, Yeast that catalyzes the incorporation of 1 nmol of AMP into RNA in 10 minutes at 37°C.

5X POLY(A) POLYMERASE, YEAST REACTION BUFFER: 250 mM Tris-HCl, 1 M NaCl, 50 mM $MgCl_2$, 5 mM DTT, pH 8.0

HEAT INACTIVATION: 65°C for 20 minutes

PROTOCOL:

3'-End labelling of RNA:

1. Add the following components to a sterile tube sitting on ice:

Components	Volume	Final Concentration
RNA	Variable	0.2 µM
Cordycepin-5'-Triphosphate	Variable	0.4 µM
Poly(A) Polymerase, Yeast (1 U/µl)	1 µl	1 µl
5X Poly(A) Polymerase, Yeast Reaction Buffer	2 µl	1X
25 mM $MnCl_2$	1 µl	2.5 mM
Nuclease-free H_2O	upto 10 µl	-

2. Collect all components by a brief centrifugation. Incubate the reaction at 37°C for 10 min.
3. The 3'-End labelled RNA product is ready for immediate downstream applications or for long-term storage at -80°C.

Poly(A) tailing of RNA:

1. Add the following components to a sterile tube sitting on ice:

Components	Volume	Final Concentration
RNA	Variable	0.2 µM
ATP (10 mM)*	1.25 µl	0.5 mM
Poly(A) Polymerase, Yeast (1 U/µl)	1 µl	1 U
5X Poly(A) Polymerase, Yeast Reaction Buffer	5 µl	1X
25 mM $MnCl_2$	2.5 µl	2.5 mM
Nuclease-free H_2O	upto 25 µl	-

**Radiolabelled, biotinylated or fluorescently-labelled ATP can be substituted in the reaction.*

2. Collect all components by a brief centrifugation. Incubate the reaction at 37°C for 10 to 20 minutes.
3. Terminate the reaction by heating at 65°C for 20 minutes or by adding 5 mM EDTA.
4. The Poly(A)-tailed RNA product is ready for immediate downstream applications or for long-term storage at -80°C.

RELATED PRODUCTS:

- Advance™ DNA Polymerase (Cat# M1151-250, -1000)
- Blood Advance™ DNA Polymerase (Cat# M1153-100, -400)
- Breeze™ DNA Polymerase (Cat# M1148-250, -1000)
- Distant™ DNA Polymerase (Cat# M1150-250, -1000)
- Fire Start™ DNA Polymerase (Cat# M1149-250, -1000)
- Outstretched™ DNA Polymerase (Cat# M1152-250, -1000)
- PFU DNA Polymerase (Cat# 9003-500, -2500)
- Ready™ DNA Polymerase (Cat# M1146-1000, -5000, -10000)
- Robust Ready™ DNA Polymerase (Cat# M1147-250, -1000)
- Taq DNA Polymerase (Cat# 9001-500, -2500)

FOR RESEARCH USE ONLY! Not to be used on humans.

BioVision Incorporated
Tel: 408-493-1800 | Fax: 408-493-1801
155 S. Milpitas Boulevard, Milpitas, CA 95035 USA

www.biovision.com | tech@biovision.com